Certificate of Analysis

Pepsin:

Part No.	Size
V195A	250mg

Description: Pepsin preferentially cleaves at the C-terminus of phenylalanine, leucine, tyrosine and tryptophan (1–4). This protease can be used alone or in combination with other proteases for protein analysis by mass spectrometry and other applications.

Biological Source: Porcine stomach.

Molecular Weight: 34.6kDa.

Form: Lyophilized.

Storage Conditions: See the Product Information Label for storage conditions and expiration date.

Optimal pH: 1.0–3.0 (4–6).

Activators: Hydrochloric acid (HCl), trifluoroacetic acid (TFA).

Inactivators: pH greater than 6.0.

Usage Notes:

1. Resuspend Pepsin in double-distilled water (pH 5.5 or lower) to a final concentration of 1mg/ml. Store reconstituted Pepsin at 4°C for up to 1 month.
2. Specificity for cleavage at Phe and Leu is best at pH 1.0 and decreased above pH 2.0. Pepsin irreversibly inactivates above pH 6.0.

Quality Control Assays

This lot passes the following Quality Control specifications:

Activity: Digestion reactions using insulin as a substrate are performed at a protease:substrate ratio of 1:20 and analyzed by reverse-phase HPLC. Intact substrate is undetectable after incubation for 15 minutes at 37°C.

Usage Information on Back

Signed by:

R. Wheeler, Quality Assurance

Part# 9PIV1959

Revised 8/16

AF9PIV19590816V1959

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Usage Information

1. In-Solution Digestion Protocol

1. Resuspend the protein in buffer at pH 7.5.
2. Transfer the protein solution to a microcentrifuge tube.
3. Add 1N HCl to the solution to a final concentration of 0.04N.
4. Resuspend Pepsin in double-distilled water.
   Note: Resuspending the enzyme in buffer at neutral or higher pH will destabilize or irreversibly inactivate it.
5. Add Pepsin to protein solution; mix. We recommend using enzyme:protein ratios of 1:20 to 1:100 (w:w).
6. Mix and centrifuge briefly.
7. Incubate 1–18 hours at 37°C.
8. Stop the reaction by heating at 95°C for 10 minutes.

2. References

1. Aiten, A. *et al.* (1989) *Protein Sequencing: A Practical Approach*, IRL Press, Oxford, UK, 43.
2. Christenses, L.K. (1955) *Arch. Biochem.* **57**, 163–73.
3. Sweeney, P.J. and Walker, J.M. (1993) In: *Enzymes of Molecular Biology*, Burrell, M.M., ed., HumanaPress, Totowa, NJ, 290–1.
4. Dixon, M. *et al.* (1979) *Enzymes*, Academic Press, New York, NY, 262.
5. Schlamowitz, M. *et al.* (1959) *J. Bio. Chem.* **234**, 3137–45.
6. Cornish-Bowden, A.J. and Knowles, J.R. (1969) *BioChem. J.* **113**, 353–62.

3. Related Products

Product	Size	Conc.	Cat.#
Asp-N, Sequencing Grade	2µg		V1621
Arg-C, Sequencing Grade	10µg		V1881
Chymotrypsin, Sequencing Grade	25µg		V1061
	100µg (4 × 25µg)		V1062
Elastase	5mg		V1891
Endo H	10,000u	500u/µl	V4871
	50,000u	500u/µl	V4875
Endoproteinase Lys-C, Sequencing Grade	5µg		V1071
Fetuin	500µg	10mg/ml	V4961
Glu-C, Sequencing Grade	50µg (5 × 10µg)		V1651
Immobilized Trypsin	2ml		V9012
	4ml (2 × 2ml)		V9013
PNGase F	500u	10u/µl	V4831
ProteaseMAX™ Surfactant, Trypsin Enhancer	1mg		V2071
	5 × 1mg		V2072
Protein Deglycosylation Mix	20 reactions		V4931
rLys-C, Mass Spec Grade	15µg		V1671
Sequencing Grade Modified Trypsin	100µg (5 × 20µg)		V5111
Sequencing Grade Modified Trypsin, Frozen	100µg (5 × 20µg)		V5113
Thermolysin	25mg		V4001
Trypsin Gold, Mass Spectrometry Grade	100µg		V5280
Trypsin/Lys-C Mix, Mass Spec Grade	20µg		V5071
	100µg		V5072
	100µg (5 × 20µg)		V5073

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